

## FORMULATION AND DEVELOPMENT OF *HEMIDESMUS INDICUS* (L.) ELIXIR FOR THE TREATMENT OF RENAL CALCULI

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### ABSTRACT:

*Hemidesmus indicus* (L.) or **Sarsaparilla** belongs to the family Apocynaceae is widely used in traditional medicine for the various biological activities attributed to its different parts, especially the roots.[5] Sarsaparilla is rich in a wealth of plant components having beneficial effects on the human body. Sarsaparilla is rich in antioxidants, flavonoids, and phenolic acids and considered safe for most of the people.[2] It is used to treat kidney stones or renal calculi, Poly Cystic Ovarian Syndrome, urinary tract infections, ulcerations, asthma etc. Renal calculi have multiphase etiological factors with high recurrence rate which affects around 12% of world population. To reduce the recurrence rate, homoeopathic interventions can be the most effective alternative option and for that one of the frequently prescribed medicines is Sarsaparilla used by homoeopaths in renal stones.[4] Elixirs are clear, pleasantly flavored, sweetened hydro-alcoholic liquids intended for oral use and they are better than the aqueous syrups to maintain both water soluble and alcohol soluble components in the solutions because of their hydroalcoholic character.[6] In this present work an elixir dosage form of Sarsaparilla alcoholic extract was formulated as this drug is commonly prescribed by Homeopathic physicians for treating renal calculi. Formulations were followed by some evaluation studies e.g. stability studies, pH determinations, optical density determination, viscosity determination, surface tension determination and TLC were also carried out for confirmation of flavonoids in the prepared formulations.

**KEYWORDS:** *Sarsaparilla, Cranberry, Renal Calculi, Elixir.*

### INTRODUCTION:

*Hemidesmus indicus* (L.) or **Sarsaparilla** belong to the family Apocynaceae being widely used in traditional medicine in the different parts of the Indian subcontinent also in European medicine for the various biological activities attributed to its different parts, especially the roots.[1][5] This plant contains various phytoconstituents like flavonoids, tannins, glycoside, sterols, and volatile oil and considered safe for most of the people.[3] It is used to treat kidney stones or renal calculi,

PCOS, urinary tract infection, ulceration, asthma etc. Renal calculi have multiphase etiological factors with high recurrence rate which affects around 12% of world population. Kidney stones is

a complex disease; therefore, their occurrence, prevention, and recurrence are all difficult to predict.[3] Larger stones, especially those with a diameter of 7mm or more, are causing obstruction along their site of occurrence with a slight change in dietary and fluid intake. The most common and major type of kidney stones is calcium oxalate. Due to the dehydration, changes in the diet, changes in hormones and due to the kidney infection, the kidney stone is formed. The basic symptoms of this disease are renal pain, blood and pus in urine, fever. To reduce the recurrence rate, use of homoeopathic intervention can be the most effective alternative option and Sarsaparilla is one of the frequently prescribed medicines.[3] Though preliminary research is promising, more clinical data is necessary to validate any claim of potential health benefits of sarsaparilla.[4] Cranberry which was included in our prepared formulation, is a popular ingredient in dietary supplements and are not known to be associated with any serious risks for human health when consumed properly. Cranberry can help to reduced risk of kidney stones. It helps to reduce the pH of urine by providing an acid load decreasing the urinary uric acid by retarding urate synthesis. Elixirs are clear, pleasantly flavored, sweetened hydro-alcoholic liquids intended for oral use and they are better than the aqueous syrups to maintain both water soluble and alcohol soluble components in the solutions because of their hydroalcoholic character.[6] In this present work an elixir dosage form of Sarsaparilla alcoholic extract is formulated as this drug is commonly prescribed by Homeopathic physicians for treating renal calculi followed by some evaluation studies e.g. stability study, pH stability, optical density and percentage stability study, clarity study, TLC, viscosity and surface tension study.[4] Another key ingredient of this formulation is cranberry which is also beneficial for the treatment of renal calculi. A red-coloured clear overall stabled elixir is formulated which can dissolve the calcium oxalates primarily which is the main composition of renal calculi.

Kidney stone is an accumulative urological disorder of human health which is a crystal concretion generally formed within the kidneys, affecting about 12% of the world population. The most common type of kidney stone is Calcium Oxalate, which forms at Randall's plaque on the renal papillary surfaces and highly prevalent with rates of up to 14.8% and increasing. The recurrence rate of the formation of the stone is up to 50% within the first 5 years of the initial incident. The presence of kidney stones is an important and challenging clinical problem. Numerous physiochemical events such as, super saturation, nucleation, growth, aggregation, and retention of urinary stone constituents within tubular cells are responsible for the formation of the stone making it a complex process. Several therapies including thiazide diuretics, allopurinol, painkillers, dietary changes, shock-wave treatment, ureteroscopic and percutaneous nephrolithotomy, and ureteroscopic and percutaneous nephrolithotomy in severe cases are being performed for treatment of kidney stone.[3] To help in preventing stones medical therapy, in conjunction with dietary measures, can be used. But these treatments possess multiple side effects and reoccurrence which have motivated researchers to search for better and safer options. Several medicinal plants such as Sarsaparilla perform an important role in treatment of renal calculi due to their antiurolithiatic activity. Though the Sarsaparilla is offering wonderful results in cases of urolithiasis in a very non-invasive and economical way without surgery but still it is not yet popularized. It is one of the best alternatives for those people who are afraid of surgeries.[4] Dehydration, changes in the diet, changes in hormones and the kidney infection causes the occurrence of kidney stones, showing the symptoms like renal pain, blood in urine, pus in urine, and fever. [8] In this disease one of the most important factors is the size of the kidney stone. Human body can pass smaller kidney stones naturally through urination before causing any

symptoms and may not require any treatment but in cases of larger stones, where symptoms are seen, to alleviate the symptoms and to get rid of the stone's treatment is recommended. [9] These larger size of kidney stones can block urine flow and cause painful symptoms such as:

- Feeling of urgency, frequent, painful, burning urination as well as difficulty in urination
- Pain in lower back and the abdomen
- Urinary retention
- Blood in urine
- Pus in urine
- Fever
- in case of infection nausea, vomiting, chills and fever can be seen
- Pain worsens during movement

These renal calculi differ in size, shape and chemical compositions depends on the chemical imbalance in the urine composition.[7] Renal calculus is generally classified into following types, based on differences in mineral composition [8] and pathogenesis, - 1) Calcium stones, 2) Struvite or Magnesium Ammonium Phosphate Stones, 3) Uric Acid Stones or Urates, 4) Cystine Stones, 5) Drug-Induced Stones 6) Cholesterol stones and 7) Lecithin kidney stones.

Elixirs are a liquid oral dosage form and are composed of sweetened hydroalcoholic liquids. Typically, when the drug is not dissolved in water alone, alcohol and water are used as solvents. To improve patient acceptance, in addition to the active drug, they usually contain flavouring and colouring agents.[6]

*H. indicus*, from the family Apocynaceae, commonly known as Anantamul.[5] The roots of *H. indicus* have been used as tonics, diuretics, and alternatives. *H. indicus* has been used as a traditional remedy for kidney diseases, jaundice, fever, cough, headache, pain, inflammation, dysentery and diarrhoea, infections, skin disease, menorrhagia, post-partum recovery, nephritic complaints, diabetes, stomach-ache, gastrointestinal disorders, edema, mouth sores, toothache, gonorrhoea, syphilis, impotence, snakebite, scorpion sting, leucoderma, psoriasis, and rheumatism. It is also being used as a blood purifier, body coolant, appetizer, and health and vitality promoter.[1]

Sarsaparilla has a potential role as an inhibitor of calcium oxalate and calcium phosphate crystallisation which can use to treat the renal calculi and it is also effective to inhibit the recurrent rate of it. The compounds in sarsaparilla that are believed to contribute to its diuretic properties include saponins and flavonoids. Saponins have been suggested to increase urine production, which could potentially help flush out kidney stones. Flavonoids, on the other hand, may help relax smooth muscles and improve blood flow, which could aid in the passage of kidney stones. [4]

Fig: sarsaparilla root



## OBJECTIVES:

- ✓ To formulate and evaluate the sarsaparilla elixir.

- ✓ To optimize the formulation.
- ✓ To perform overall stability studies.
- ✓ To develop taste masking of the formula (alcoholic extract).
- ✓ To carry out In-vitro testing and design an evaluation set up.
- ✓ To study the formulations in clinical subjects.

## MATERIALS AND METHODS:

### Materials:

Sarsaparilla alcoholic extract (Sett Dey & Co. Lab), Cranberry extract (Manama company), Sodium citrate (Research grade), Sodium metabisulphite (Research grade), Di sodium EDTA (Research grade), Sodium Benzoate (Research grade), Glycerol (Research grade), Stevia Powder (Research grade).

### Instruments Used:

Lab Glasswares (Borosilicate), Brookfield Viscometer (LV), pH meter (Elico), Photoelectric colorimeter (Systronics), Stability Chamber (Labard), TLC chamber (Borosilicate).

### Method Of Preparation:

- Cranberry extract (pulpy syrup) was clarified & filtered.
- The filtrate was collected.
- Different trial batches were prepared (F1 to F4).
- Sarsaparilla alcoholic extract (drug) and excipients (Sodium citrate, Sodium metabisulphite, Di sodium EDTA, Sodium Benzoate, Glycerol, Stevia Powder) were mixed in different ratio to formulate different trial batches.



**Fig: Sarsaparilla Elixir (trial batch F1 to F4)**

### Formulation Chart:

SL. NO.	INGREDIENTS	F1	F2	F3	F4
1	Sarsaparilla Alcoholic Extract	10ml	15ml	20ml	25ml
2	Cranberry Extract	89ml	84ml	79ml	74ml
3	Sodium Citrate	0.1gm	0.1gm	0.1gm	0.1gm
4	Sodium Metabisulphite	0.1gm	0.1gm	0.1gm	0.1gm
5	Sodium EDTA	0.1gm	0.1gm	0.1gm	0.1gm
6	Sodium Benzoate	0.1gm	0.1gm	0.1gm	0.1gm
7	Glycerol	1ml	1ml	1ml	1ml
8	Stevia Powder	q.s.	q.s.	q.s.	q.s.

**TABLE 1: Formulation of Sarsaparilla Elixir (Trial Batches)**

### EVALUATIONS:

Basic evaluation of the Sarsaparilla elixir is performed after the development of the formula, which are listed below-

<u>Sl no.</u>	<u>Evaluation study (name)</u>	<u>Equipment name</u>	<u>Purpose</u>	<u>Remark</u>
1	Overall stability study	Room temperature	Stability of formulation	Stable formula
2	pH determination	digital pH meter	pH study of formulation	Initially acceptable
3	Colour Index determination	colorimeter	% transmittance, optical density of the formulation	Initially acceptable
4	Viscosity	Brookfield Viscometer	Viscosity study of the formulation	Initially acceptable
5	Surface tension	stalagmometer	Surface tension study of the formulation	Initially acceptable
6	Accelerated stability study	stability chamber	stability study of the formulation	Stable formula

7	Freeze drying	freeze dryer	stability study of the formulation	Stable formula
8	TLC	TLC chamber	Quality study of the formulation	Initially acceptable Standard formulation

**TABLE 2: Evaluation study of Sarsaparilla Elixir (Trial Batches)**

**RESULTS AND DISCUSSIONS:**

- An overall stable formulation of Sarsaparilla elixir was prepared, primarily which dissolved calcium oxalate and phosphate successfully.
- The formulation was a rosy red coloured clear elixir containing Sarsaparilla alcoholic extract as alcoholic part and Cranberry as aqueous part to treat the renal calculi and prevent the recurrent rate of renal calculi.

**Overall Stability Study:** The prepared formulation of Sarsaparilla elixir was kept in glass container at room temperature and Stability chamber for 6 months and was evaluated periodically. The formulations were found to be stable during the study.

**pH Determination:** The pH of the formulations of the trial batches were determined using digital pH meter. The pH range was 5 to 6.2.

SL NO	F1	F2	F3	F4
1	6.2	5.8	5.3	5
2	6.2	5.8	5.3	5
3	6.2	5.8	5.3	5
4	6.2	5.8	5.3	5
5	6.2	5.8	5.3	5



**TABLE 3: pH Determination of Sarsaparilla Elixir Trial Batches**

**Fig: digital pH meter**

**Colour Index Determination:** The percentage transmittance and optical density of each batch was determined using colorimeter at filter 2 which shows the stability of the batches.



**Fig: Photoelectric colorimeter**

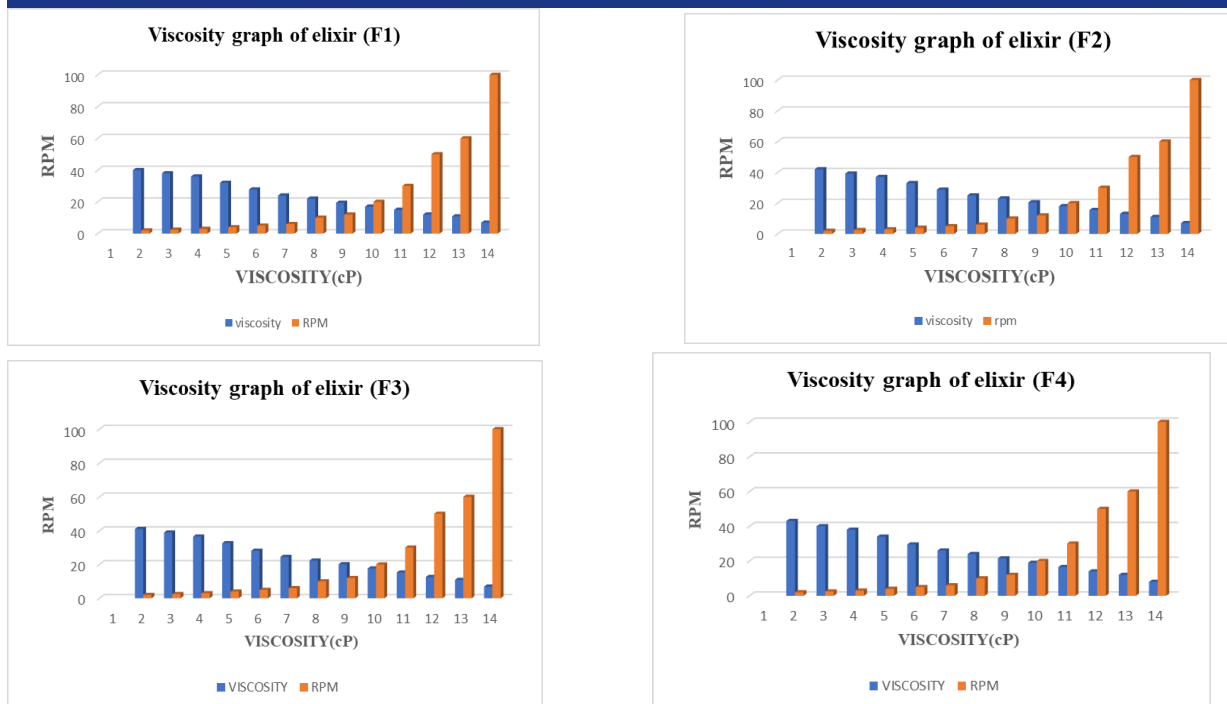
SL NO	FILTER 2	F1	F2	F3	F4
1	%T	25	26	28	28
	OD	0.61	0.63	0.84	0.75
2	%T	23	26	28	28
	OD	0.64	0.63	0.80	0.75
3	%T	23	25	28	28
	OD	0.64	0.63	0.80	0.75
4	%T	23	23	27	28
	OD	0.64	0.63	0.81	0.75
5	%T	23	23	28	28
	OD	0.64	0.63	0.80	0.75

**TABLE 4: Colour Index Study of Sarsaparilla Elixir Trial Batches**

**Viscosity:** Viscosity measurement is one of the most important quality control tests for *elixirs*. The consistency of elixir should be clear it was determined by using Brookfield viscometer. Formulations were slightly viscous with acceptable consistency. The spindle number S61 is used here for the evaluation study. Average viscosity of sarsaparilla elixir is 23.97cP.

Sl. No.	Sample 1(F1)		Sample 2 (F2)		Sample 3 (F3)		Sample 4 (F4)	
	viscosity	rpm	viscosity	rpm	viscosity	rpm	viscosity	rpm
1	40	2	42	2	41	2	43	2
2	38	2.5	39.2	2.5	38.9	2.5	40	2.5
3	36	3	37	3	36.5	3	38	3
4	32	4	33	4	32.6	4	34	4
5	27.8	5	28.75	5	28.12	5	29.5	5
6	24	6	25	6	24.5	6	26	6
7	22	10	23	10	22.3	10	24	10
8	19.43	12	20.5	12	20.1	12	21.5	12
9	17	20	18	20	17.65	20	19	20
10	15	30	15.5	30	15.25	30	16.5	30
11	12	50	13	50	12.6	50	14	50
12	10.86	60	11	60	10.9	60	12	60
13	6.89	100	7	100	6.9	100	8	100

**TABLE 5: Viscosity study of Sarsaparilla Elixir Trial Batches (F1- F4) using Brookfield viscometer**



**Fig1: Graphical representation of viscosity of elixir trial batches using Brookfield Viscometer**

**Surface Tension:** It is well known that surface tension is considered as a critical parameter for oral liquid dosage forms that is often overlooked since they are quickly distributed in the physiological aqueous media of the body. this knowledge may have a greater impact on efficiency of the delivery of the drug and in the determination of the  $pK_a$  value of drugs. Surface tension is derived from a liquid’s elastic tendency and helps to determine significant liquid–air exposures by determining the layer of molecules on the surface attempt to minimize their overall surface area in the bulk liquid by being attracted to molecules. Here, surface tension of the elixir is determined using stalagmometer by drop count method.

The average of surface tension of the trial batches of Sarsaparilla elixir was 63.8 dynes/cm which is in the acceptable range of 36.6–64.7 dynes/cm for oral liquid dosage form. [11]



**Fig: stalagmometer**

SL NO	Sample 1 (F1)		Sample 2 (F2)		Sample 3 (F3)		Sample 4 (F4)	
	Weight (g)	Surface tension ( $\gamma$ )	Weight (g)	Surface tension ( $\gamma$ )	Weight (g)	Surface tension ( $\gamma$ )	Weight (g)	Surface tension ( $\gamma$ )
1	22.06	63	21.97	62.2	22.25	64.6	22.35	65.4

**TABLE 6: Surface tension study of Sarsaparilla Elixir Trial Batches (F1- F4)**



**Accelerated Stability Study:** Accelerated stability chambers are used for accelerated aging studies. This type of testing allows pharmaceutical companies to test long-term stability in a short-term time frame by involving subjected drugs to extreme environmental conditions such as high temperatures, humidity levels, and light exposure levels etc. Stability chambers are essential for ensuring product safety, quality by testing the effects of various environmental factors on drug performance. Samples were studied using the Stability chamber by following standard protocols. The stability profile was satisfactory enough.



**Fig: Sarsaparilla elixir sample**

**After 4 weeks the sample showed overall stability which is initially acceptable.**

NAME OF TEST	F1	F2	F3	F4	REMARK
Surface tension	63	62.2	64.6	65.4	Initially acceptable
Viscosity	23.15	24.07	23.64	25.03	Initially acceptable
pH	6.2	5.8	5.3	5	Initially acceptable
C.I.	No changes	No changes	No changes	No changes	Initially acceptable
Phase separation	No separation	No separation	No separation	No separation	Initially acceptable

**After 8 weeks the sample showed overall stability which is initially acceptable.**

NAME OF TEST	F1	F2	F3	F4	REMARK
Surface tension	63	62.2	64.6	65.4	Initially acceptable
Viscosity	23.15	24.07	23.64	25.03	Initially acceptable
pH	6.2	5.8	5.3	5	Initially acceptable
C.I.	No changes	No changes	No changes	No changes	Initially acceptable
Phase separation	No separation	No separation	No separation	No separation	Initially acceptable

**After 12 weeks the sample showed overall stability which is initially acceptable.**

NAME OF TEST	F1	F2	F3	F4	REMARK
Surface tension	63	62.2	64.6	65.4	Initially acceptable
Viscosity	23.15	24.07	23.64	25.03	Initially acceptable
pH	6.2	5.8	5.3	5	Initially acceptable
C.I.	No changes	No changes	No changes	No changes	Initially acceptable
Phase separation	No separation	No separation	No separation	No separation	Initially acceptable

**TLC:** Thin layer chromatography (TLC) is an analytical technique often used to separate and identify compounds present in a given mixture. It can also be used to determine the purity of a particular substance within that mixture.

Here, TLC was performed to determine the quality of the drug in the formulation by comparing the API with standard marketed formulation of Sarsaparilla using a TLC chamber. To perform the test a TLC plate was prepared using Silica gel G. The solvent was prepared using chloroform: glacial acetic acid: methanol: water at the ratio of 16:8:3:2. The R<sub>f</sub> values of the sample and standard was same which shows the quality of the sample was same as standard of Sarsaparilla used in the formulation.

R<sub>f</sub> = (distance travelled by the solute / distance travelled by the solvent)

R<sub>f</sub> 1 = 6.45/8 = 0.80 (for standard and sample)

R<sub>f</sub> 2 = 7.04/8 = 0.88 (for standard and sample)

R<sub>f</sub> 3 = 7.2/8.18 = 0.88 (for standard and sample)

R<sub>f</sub> 4 = 6.25/7.81 = 0.80 (for standard and sample)

Brilliant white coloured spots having the average R<sub>f</sub> value of 0.84 is detected under the UV light which is within the acceptable range of standard powder of root of Sarsaparilla. (0.13 to 0.88)



**Fig: TLC plate**

SL No.	TLC Plate 1 (F1)		TLC Plate 2 (F2)		TLC Plate 3 (F3)		TLC Plate 4 (F4)	
	Standard R <sub>f</sub> 1	Sample R <sub>f</sub> 1	Standard R <sub>f</sub> 2	Sample R <sub>f</sub> 2	Standard R <sub>f</sub> 3	Sample R <sub>f</sub> 3	Standard R <sub>f</sub> 4	Sample R <sub>f</sub> 4
1	0.80	0.80	0.88	0.88	0.88	0.88	0.80	0.80

**TABLE 5: TLC study of Sarsaparilla Elixir Trial Batches (F1- F4)**

R<sub>f</sub> avg. = 0.84, within the acceptable range of 0.13 to 0.88 for standard commercially available powder of root of Sarsaparilla. [10]

## CONCLUSION:

We were able to formulate a stable Elixir formulation of *Hemidesmus indicus* (L.) for the Treatment of Renal Calculi. An overall stable clear red coloured elixir was prepared using Sarsaparilla as active drug constituent to treat the renal calculi which primarily dissolved calcium oxalate and phosphate successfully. Thus, the present study was a successful formulation in terms of the laid down objectives

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